**High-throughput glycomic analysis of different mammalian antibodies as a strategy in laboratory diagnosis and identification of disease biomarkers**

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Using affinity chromatography by use of monolithic supports with different immobilized ligands that bind different mammalian antibodies, a robust tool for high-throughput isolation (HTP) of different immunoglobulins, such as IgG, IgA and from serum and cell supernatants was developed. Compared to proteins A and G, recombinant protein L binds by far the largest number of isoforms of all immunoglobulins. For this reason, this ligand immobilized on a monolithic column was mostly used. For isolation of highly enriched IgG, IgA and IgM preparations, the fraction with proteins eluted from the affinity column was further separated by ion-exchange chromatography followed by 1D polyacrylamide gel electrophoresis. As final step in sample preparation, in-gel tryptic digestion of separated proteins protein was performed. The resulting belonging peptides to heavy and light IgG, IgA and IgM chains were successfully identified by MALDI-ToF/ToF mass spectrometry. After isolation of immunoglobulins, their glycans have been removed from the protein chain using deglycosylation enzyme PNGase F. The IgG and IgM glycan structures of different mammalian species were analysed by use of hydrophilic interaction chromatography (HILIC) followed by identification by use of MALDI-ToF/ToF MS. Presented results show differences between glycan structures of immunoglobulins isolated from different mammalian species (human, mouse and rat), as well as changes in glycan structures of human and rat immunoglobulins during disease development and during treatment. Possible use of this strategy for laboratory diagnosis and identification of disease biomarkers is presented.